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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
09/748,690	12/22/2000	Melis Anastasios	NREL 99-29	4270

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PAUL J WHITE, SENIOR COUNSEL
NATIONAL RENEWABLE ENERGY LABORATORY (NREL)
1617 COLE BOULEVARD
GOLDEN, CO 80401-3393

EXAMINER
AFREMOVA, VERA

ART UNIT	PAPER NUMBER
1651	

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14

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary

Application No.	Applicant(s)
09/748,690	Melis Anastasios
Examiner	**Art Unit**
Vera Afremova	1651

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136 (a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1) ☒ Responsive to communication(s) filed on *Feb 6, 2003*.

2a) ☐ This action is **FINAL.** 2b) ☒ This action is non-final.

3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

Disposition of Claims

4) ☒ Claim(s) *1-3, 5-8, and 10* is/are pending in the application.

4a) Of the above, claim(s) _____ is/are withdrawn from consideration.

5) ☐ Claim(s) _____ is/are allowed.

6) ☒ Claim(s) *1-3, 5-8, and 10* is/are rejected.

7) ☐ Claim(s) _____ is/are objected to.

8) ☐ Claims _____ are subject to restriction and/or election requirement.

Application Papers

9) ☐ The specification is objected to by the Examiner.

10) ☐ The drawing(s) filed on _____ is/are a) ☐ accepted or b) ☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11) ☐ The proposed drawing correction filed on _____ is: a) ☐ approved b) ☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12) ☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13) ☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a) ☐ All b) ☐ Some* c) ☐ None of:

1. ☐ Certified copies of the priority documents have been received.
2. ☐ Certified copies of the priority documents have been received in Application No. _____.
3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

*See the attached detailed Office action for a list of the certified copies not received.

14) ☒ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

a) ☐ The translation of the foreign language provisional application has been received.

15) ☐ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s). _____
4) ☐ Interview Summary (PTO-413) Paper No(s). _____
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other:

DETAILED ACTION

Continued Examination Under 37 CFR 1.114

A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicants' submission filed on 2/06/2003 has been entered.

Status of claims

Claims 1-3, 5-8 and 10 as amended are under examination in the instant office action [Paper No. 13 filed 2/06/2003].

Claims 4 and 9 were canceled by applicants [Paper No. 6 filed 5/28/2002].

Claim Objections

Claims 1-3, 5-8 and 10 as amended are objected to because of the following informalities:

There are some typing errors in the claims. For example: phrase "microorganism" in claims 1, line 3; phrase "becomes" as related to several "cells" in claim 1, line 7; article "a" as related to plural form of phrase "algae" in claim 6; coma before "and" in claim 6, line 2. Appropriate corrections are required.

Response to Arguments

Applicants' arguments filed 2/06/2003 have been fully considered but they are not all found persuasive for the reasons below.

Claim Rejections - 35 USC § 112

Claims 1-3, 5-8 and 10 as amended remain/are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention for the reasons as explained in the prior office action and for the reasons as explained below.

The claimed method as amended remains indefinite with regard to the phrase "**temporal** separation of oxygen evolution" because it is uncertain when and from what "oxygen evolution" is separated. Is it from hydrogen ("hydrogen production") or from carbon dioxide ("rate of cellular respiration)? Or whether is "separation" achieved by "sealing the culture from atmospheric oxygen" (step b) ? Is it complete separation in time? What are time periods?

The claimed method as amended remains indefinite with regard to step of "depleting a nutrient". It is unclear whether the nutrient was present in a medium of step (a), whether the nutrient is used (depleted or reduced) during microbial grow to a concentration less that 0.5 millimolar (see claim 3) or whether the medium is replaced by another medium without the claimed nutrient(s). It is uncertain as claimed whether a depleting step is an active step such as changing one nutrient medium for another medium or whether a depleting step is an inherent process as the result of growing microorganisms and, thus, using/depleting/reducing available nutrients in the medium. It appears from the as-filed specification that the culture has been deprived from sulfur by replacing the first culture medium with the second sulfur-free medium (page 5, last 3 lines and/or page 8, last par.). Yet, the claimed invention does not appear to point

out the active step of depriving or removing selected nutrient as intended and/or argued by applicants, for example: see response page 4, last paragraph.

Claim 1 remains indefinite and confusing with regard to the use of light energy during the whole process because it is not particularly clear whether or not some differences are intended between "illuminated conditions" in step (a) and "light" condition in the instant step (e). Claim 1 as amended adds to the confusion because it is now uncertain whether "the light of saturating yellow actinic excitation", which is applied to the sample in step (d), is also intended for the whole process including steps (a), (b) and (e).

Claim 1 as amended is rendered indefinite by the phrase "controlling" because it is unclear what is done in this step (e) in order to control rates of oxygen production and of respiration as intended. The claimed method encompasses some measurements in the samples drawn from the whole culture but it is unclear whether the results of steps (c) and (d) have been evaluated to control events of the step (e) as claimed.

Claim 2 recites the limitation "**a** hydrogenase" in the process of claim 1. The process of claim 1 refers two times to this enzyme, for example: "hydrogenase" (see preamble) and "reversible hydrogenase through photosynthesis" (see step (e)). Thus, the antecedent basis for this limitation becomes confusing and uncertain as claimed. Please, amend both claims to provide a clear antecedent basis for hydrogenase as intended.

Claim 2 is not particularly clear whether hydrogen is generated from both water and from "endogenous substrate". Please, provide clarification and insert "from" second time is that is intended.

Claim 5 remains indefinite with respect to the limitation "a plurality of cycles". The pending claim 5 appears to refer to the method of the original claim 1 but not to the amended claim 1 and, thus, the steps recited in the claim 5 lack proper and sufficient antecedent basis in the amended claim 1. It is uncertain what steps are repeated for a plurality of cycles.

Claim 7 as now pending appears to refer to the method of the original claim 1 but not to the amended claim 1 and, thus, the active steps recited in the claim 7 lack proper and sufficient antecedent basis in the amended claim 1. For example: the phrase "generating" clearly lacks antecedent basis and it is uncertain whether the step of "incubating" is directed to growing and/or incubating the sample or the whole culture.

Claim 8 recites "substrate". It is not particularly clear as claimed whether "substrate" of claim 8 is "endogenous substrate" for generation of hydrogen or whether "substrate" of claim 8 might be substrate in a culture medium.

New matter

Claims 1-3, 5-8 and 10 as amended are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

Insertion of the limitation drawn to "controlling" rates of oxygen production and of cellular respiration in step (e) raises the issue of new matter with respect to the proper support in the as-filed specification.

First, the insertion of this limitation is a new concept because it does not have a literal support in the as-filed specification by way of a generic disclosure of "controlling" as an active step as presently claimed.

Further, this newly inserted limitation appears to encompass the active step of "controlling" rate of oxygen production to be equal or less than respiration in the whole culture after the rate of oxygen production and the rate of respiration are measured in the samples drawn from the whole culture system. There are some particular examples in as-filed specification which demonstrate how oxygen production and respiration were measured in some samples (page 6). But there is no specific examples in the as-filed specification which demonstrate how these measurements are evaluated and/or how the results of "measuring" are applied to the whole culture in order to control the rate of oxygen production to be equal or less than respiration in the whole culture. This is a matter of written description, not a question of what one of skill in the art would or would not have known. The material within the four corners of the as-filed specification must lead to the generic concept. If it does not, the newly inserted limitation of "controlling" the rates to be equal or less as claimed raises a question of a new matter.

Applicants appear to argue that in order to avoid deactivation of hydrogenase in the presence of oxygen the fully grown algal culture has been deprived from sulfur (response pages

4-5). Thus, the actual removal of sulfur results in the unexpected metabolic switch which over some period of incubation of the algal culture, for example: 24-30 hours (see specification page 8, last par.) leads to the respiration rate being higher than the oxygen production rate. However, the presently claimed method appears to encompass the results of the "measuring" steps as the basis for the active control or "controlling" the metabolic rates in the whole physiological process.

Claim Rejections - 35 USC § 102

The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action.

The claim rejection under 35 U.S.C. 102(b) as being anticipated by US 4,010,076 [A] was withdrawn in the prior office action because the claimed method requires culturing of a photosynthetic algal microorganism. But US'076 provides the exemplified disclosure with regard to photosynthetic bacterial microorganism. However, the cited patent suggests the similar method for a variety of photosynthetic algal culture in "spent" cultures and thus, it teaches the use of a culture system with depleted nutrients or with nutrients reduced over the period of incubation for the purpose of "temporal separation" of oxygen evolution and for production of hydrogen as intended for the presently claimed method.

The claim rejection under 35 U.S.C. 102(b) as being anticipated by US 4,442,211 [IDS-1] has been withdrawn in the instant office action because the cited patent does not disclose active

steps of "measuring" and "controlling" the rates oxygen production and respiration as required by the presently amended method.

Claim Rejections - 35 USC § 103

The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action.

The claim rejection over the reference by Melis [IDS-2-1] is withdrawn in the light of affidavit under 37 CFR 1.1313 filed 11/04/2002. However, the removal of this reference does not change the core of the claim rejection under 35 U.S.C. 103(a) as explained in the prior office action and for the reasons below.

Claims 1-3, 5-8 and 10 as amended remain rejected under 35 U.S.C. 103(a) as being unpatentable over US 4,442,211 [IDS-1] and US 4,010,076 [A] taken with Wykoff et al. [U].

Claims are directed to a process of a temporal separation of oxygen evolution and hydrogen production by photosynthetic algal microorganism wherein the process comprises steps of growing the algal microorganism in a medium under illuminated conditions in order to accumulate endogenous substrate, depleting (reducing) nutrients including sulfur, iron or manganese from the medium, sealing the microorganism from atmospheric oxygen, incubating the sealed microorganism under illuminated conditions and collecting gaseous products including

hydrogen. Some claims are further drawn to hydrogen generation from water and accumulated substrate under illuminated conditions. Some claims are further drawn to a repetition of process or to a plurality of cycles. Some claims are further drawn to accumulation of endogenous substrates such as acetate or carbohydrate or proteins. Some claims are further drawn to the use of algal culture such as *Chlamydomonas reinhardtii.*

The cited references are relied upon as explained in the prior office action and repeated herein.

US 4,442,211 [IDS-1] discloses a process of a "temporal separation of oxygen evolution" and hydrogen production by a photosynthetic algal microorganism such as *Chlamydomonas reinhardtii* wherein the process comprises steps of growing the microorganism in a medium under illuminated conditions in order to accumulate endogenous substrate and, thus, depleting nutrients including sulfur, iron or manganese from the original medium by growing algal cells and by accumulating endogenous substrates. The cited method also comprises step of sealing the microorganism from atmospheric oxygen and/or incubating the microorganism under illuminated conditions or in the light in the environment free from atmospheric oxygen and carbon dioxide by passing inert gas through the culture system. The cited method comprises step of collecting gaseous products including hydrogen (col. 1 lines 62-65). The disclosed method teaches hydrogen generation by algal culture of *Chlamydomonas reinhardtii* from water and accumulated substrates under illuminated conditions (col. 1, lines 60-68 and col. 2, lines 1-3). The cited patent also encompasses a repetition of the process steps or a plurality of cycles (col. 3, lines 32-45).

Although, the method of the cited patent is not particularly clear whether sulfur, iron or manganese in the medium were depleted to the concentration of 0.5 millimolar or less as required by the presently claimed method (see claim 3), it is reasonably expected that the growing step results in the substantial depletion or in the substantial reduction of these inorganic nutrients particularly in view that the starting or the original culture medium contains these inorganic compounds as "trace elements" or in amounts less than 0.5 millimolar (col. 2, lines 54-55). Although, the method of the cited patent does not disclose active steps of "measuring" rates of oxygen production and respiration, the whole process occurs under the same conditions including exclusion of atmospheric oxygen, anaerobic conditions, culture illumination, depletion or reduction of selected nutrients and the process of the cited patent results in the production of hydrogen. Thus, the method of the cited patent is substantially similar to the presently claimed method.

The cited patent US 4,010,076 [A] is relied upon for the teaching of a process for hydrogen production by various photosynthetic algal cultures including blue, green and red algal cultures which comprises generating of hydrogen from water in the "spent" microbial cultures or in the culture media which is depleted from nutrients over some period of incubation (table 1). The cited patent discloses a process for generating hydrogen in the absence of atmospheric oxygen under illumined conditions in the "spent" culture with reduced nutrients by utilizing bacterial cells (example 1) as required by the presently claimed method. The cited patent also

teaches the value of microbial hydrogenase for producing useful products including hydrogen (col. 1, line 17).

Thus, the methods for hydrogen production of the cited patents US 4,442,211 and US 4,010,076 are substantially similar to the presently claimed method. However, the cited patents US 4,442,211 and US 4,010,076 are silent with regard to the active and/or controlled depletion or removal of selected nutrients including sulfur from the culture medium for controlled induction of a "temporal separation of oxygen evolution" and hydrogen production by algal microorganisms.

But the cited reference by Wykoff et al. [U] demonstrates that decline of oxygen production by photosynthetic microorganisms is induced by nutrient starvation and that the photosynthetic evolution of oxygen in the algal culture of *Chlamydomonas reinhardtii* is induced by sulfur starvation or removal of sulfur from the culture medium (abstract).

Therefore, it would have been obvious to one having ordinary skill in the art at the time the claimed invention was made to modify the prior art methods of the cited patents US 4,442,211 and US 4,010,076 by introducing an active step directed to controlling the nutrient depletion or sulfur starvation with a reasonable expectation of success in decreasing oxygen production and, thus, avoiding deactivation of hydrogenase in the presence of oxygen for the benefit in hydrogen generation as intended for the presently claimed invention because the prior art teaches that depletion from the culture medium of sulfur results in the decline of photosynthetic rate of oxygen production {Wykoff et al. [U]} and because it is known that

hydrogen is produced from water under anaerobic illuminated conditions by various photometabolically active algal microorganisms including red, green and blue green algae {US 4,442,211 [IDS-1], US 4,010,076 [A]} by action of hydrogenase {US 4,010,076}. One of skill in the art would have been motivated to separate oxygen and hydrogen production for the expected benefits of maximizing the production of hydrogen with the hydrogenase containing microbial cultures including various algal cultures. Thus, the claimed invention as a whole was clearly prima facie obvious, especially in the absence of evidence to the contrary.

The claimed subject matter fails to patentably distinguish over the state art as represented be the cited references. Therefore, the claims are properly rejected under 35 USC § 103.

The main applicants' argument is drawn to the idea of unexpected metabolic switch after removal of sulfur from the culture (see response page 4-5). Yet, the claimed method does not clearly indicate how the selected nutrients including sulfur are removed from the growing culture system. For example: the method of the cited patent US 4,010,076 encompasses the use of "spent" culture wherein the nutrients including sulfur have been used for microbial grow and, thus, the method comprises a step of depleting nutrients within the scope of the claimed invention. The method for temporal separation of oxygen evolution and hydrogen production of the cited US 4,442,211 comprises step of growing algal culture and, thus, using/depleting nutrients including sulfur as required by the claimed invention, wherein the metabolic switch that is argued is an inherent mechanism of action under identical condition.

Some applicants' arguments are directed to the idea that the cited patent US 4,442,211 (Greenbaum) does not recognize the problem of deactivation of hydrogenase in the presence of oxygen (page 6). However, this argument is directed to the mechanism of action of the hydrogen production by microbial culture rather than to the structural differences in the method protocols and in the active steps. Thus, the cited method which comprises the similar, if not identical, steps and/or structural elements as the claimed method, is also substantially similar in the final results as intended such as "temporal separation of oxygen evolution" and hydrogen production and, thus, it provides for avoiding a deactivation of hydrogenase as intended and/or argued.

With regard to the cited US 4,010,076 (Weetall) and the reference by Wykoff et al. applicants' main argument is directed to the concept of a mechanism of hydrogen production by algal culture rather than to the differences in the protocols of active steps (response pages 7-8). However, the method of US 4,010,076 (Weetall) comprises the similar, if not the same, steps and structural elements as the claimed method and, thus, it is reasonably expected to include the same mechanism or conditions for hydrogen production or the same "temporal separation of oxygen evolution" and hydrogen production which provides for avoiding a deactivation of hydrogenase as intended and/or argued. Moreover, the reference by Wykoff et al teaches decreasing oxygen production or separating oxygen production by decreasing amounts of nutrients such as sulfur. Therefore, the cited prior art discloses the same process as the applicants' process regardless whether or not the precise mechanism of action is recognized in the prior art.

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Vera Afremova whose telephone number is (703) 308-9351. The examiner can normally be reached on Monday to Friday from 9:00 to 5:30.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Michael Wityshyn, can be reached on (703) 308-4743. The fax phone number for this Group is (703) 308-4242.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the Group receptionist whose telephone number is (703) 308-0196.

Vera Afremova

Art Unit 1651 VERA AFREMOVA

April 18, 2003 PATENT EXAMINER

V. Afrem